

RECEIVED
CENTRAL FAX CENTERATTORNEY DOCKET NO. 9530.3
Application Serial No. 10/049,727
Page 3

FEB 26 2007

IN THE CLAIMS

Please amend the claims as follows. This listing replaces all prior versions.

1. (Cancelled).
2. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:
 - (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;
 - (ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and
 - (iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample. The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:
 - (a) contacting the sample either simultaneously or stepwise with a first antibody which binds selectively to PAI-1 and a labelled second antibody which binds selectively to multimeric vitronectin; and
 - (b) determining the second antibody bound to the complex to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.
3. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:
 - (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;
 - (ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and
 - (iii) determining the amount of active PAI-1 in the biological fluid by correlating the

ATTORNEY DOCKET NO. 9530.3

Application Serial No. 10/049,727

Page 4

amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

- (a) contacting the sample either simultaneously or stepwise with a first antibody which binds selectively to multimeric vitronectin and a labelled second antibody which binds selectively to PAI-1 ; and
- (b) determining the second antibody bound to the complex to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

4. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

- (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;
- (ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and
- (iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

- (a) contacting the sample either simultaneously or stepwise with a first antibody which binds selectively to PAI-1 and a labelled second antibody which binds selectively to multimeric vitronectin to form an active PAI-1/multimeric vitronectin/first antibody/second antibody complex;
- (b) separating said active PAI-1/multimeric vitronectin/first antibody/second antibody complex from the sample; and
- (c) determining the second antibody bound to the complex to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

ATTORNEY DOCKET NO. 9530.3

Application Serial No. 10/049,727

Page 5

5. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

(i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;

(ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and

(iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

a) contacting the sample either simultaneously or stepwise with a first antibody which binds selectively to multimeric vitronectin and a labelled second antibody which binds selectively to PAI-1 to form an active PAI-1/multimeric vitronectin/first antibody/second antibody complex;

(b) separating said active PAI-1/multimeric vitronectin/first antibody/second antibody complex from the sample; and

(c) determining the second antibody bound to the complex to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

6. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

(i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;

(ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and

(iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of

ATTORNEY DOCKET NO. 9530.3

Application Serial No. 10/049,727

Page 6

active PAI-1/multimeric vitronectin complex in the sample comprises:

- (a) simultaneously contacting the sample with a first antibody which binds selectively to PAI-1, the first antibody being immobilized on a solid support, and with a labelled second antibody which binds selectively to multimeric vitronectin; and
- (b) determining the second antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

7. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

- (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;
- (ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and
- (iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

- (a) contacting the sample with a first antibody which binds selectively to PAI-1, the first antibody being immobilized on a solid support;
- (b) contacting the solid support with a labelled second antibody which binds selectively to multimeric vitronectin; and
- (c) determining the second antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

8. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

- (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;

ATTORNEY DOCKET NO. 9530.3

Application Serial No. 10/049,727

Page 7

(ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and

(iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

(a) simultaneously contacting the sample with a first antibody which binds selectively to multimeric vitronectin, the first antibody being immobilized on a solid support, and with a labelled second antibody which binds selectively to PAI-1; and

(b) determining the second antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

9. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

(i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;

(ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and

(iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

(a) contacting the sample with a first antibody which binds selectively to multimeric vitronectin, the first antibody being immobilized on a solid support;

(b) contacting the solid support with a labelled second antibody which binds selectively to PAI-1; and

(c) determining the second antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

ATTORNEY DOCKET NO. 9530.3
Application Serial No. 10/049,727
Page 8

10. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

- (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;
- (ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and
- (iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:
 - (a) contacting the sample with a first antibody which binds selectively to PAI-1, the first antibody being immobilized on a solid support;
 - (b) contacting the solid support with a second antibody which binds selectively to multimeric vitronectin;
 - (c) contacting the solid support with a labelled third antibody which binds selectively to the second antibody; and
 - (d) determining the third antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

11. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

- (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;
- (ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and
- (iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric

ATTORNEY DOCKET NO. 9530.3

Application Serial No. 10/049,727

Page 9

vitronectin complex in the sample The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

- (a) contacting the sample with a first antibody which binds selectively to multimeric vitronectin, the first antibody being immobilized on a solid support;
- (b) contacting the solid support with a second antibody which binds selectively to PAI- 1;
- (c) contacting the solid support with a labelled third antibody which binds selectively to the second antibody; and
- (d) determining the third antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

12. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of

- (i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;
- (ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and
- (iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample, The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

- (a) contacting the sample, either simultaneously or stepwise, with a first antibody which binds selectively to PAI-1 and to which is attached one member of a capture pair and with a labelled second antibody which binds selectively to multimeric vitronectin to form a mixture;
- (b) contacting the mixture with a solid support on which is immobilized the other member of the capture pair; and
- (c) determining the second antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

ATTORNEY DOCKET NO. 9530.3

Application Serial No. 10/049,727

Page 10

13. (Currently amended) A method for determining active plasminogen activator inhibitor-Type 1 (PAI-1) in a biological fluid, the method comprising the steps of:

(i) providing a sample of a biological fluid selected from the group consisting of whole blood, plasma and serum;

(ii) measuring the amount of active PAI-1/multimeric vitronectin complex in the sample; and

(iii) determining the amount of active PAI-1 in the biological fluid by correlating the amount of active PAI-1 in the biological fluid to the amount of active PAI-1/multimeric vitronectin complex in the sample. The method of claim 1, wherein measuring the amount of active PAI-1/multimeric vitronectin complex in the sample comprises:

(a) contacting the sample either simultaneously or stepwise, with a first antibody which binds selectively to multimeric vitronectin and to which is attached one member of a capture pair and with a labelled second antibody which binds selectively to PAI-1 to form a mixture;

(b) contacting the mixture with a solid support on which is immobilized the other member of the capture pair; and

(c) determining the second antibody bound to the solid support to measure the amount of active PAI-1/multimeric vitronectin complex in the sample.

14-15. (Canceled).

16. (Previously presented) The method according to claim 3, wherein the second antibody is labelled with a directly detectable label.

17. (Previously presented) The method according to claim 3, wherein the second antibody is labelled with a component of a signal-generating system.

18. (Previously presented) The method of claim 17 wherein the component is an enzyme selected from the group consisting of alkaline phosphatase, amylase, luciferase, catalase,

ATTORNEY DOCKET NO. 9530.3
Application Serial No. 10/049,727
Page 11

beta-galactosidase, glucose oxidase, glucose-6-phosphate dehydrogenase, hexokinase, horseradish peroxidase, lactamase, urease and malate dehydrogenase.

19. (Previously presented) The method according to claim 3, wherein the second antibody is labelled with a fluorophore.

20. (Previously presented) The method of claim 19 wherein the fluorophore is selected from the group consisting of a coumarin, a rare earth metal ion, chelate or chelate complex, a fluorescein, rhodamine and a rhodamine derivative.

21. (Previously presented) The method of claim 3, wherein the second antibody is labelled with a luminescent material.

22. (Previously presented) The method of claim 21 wherein the luminescent material is selected from the group consisting of a cyclic diacyl hydrazide, luminol, isoluminol, an acridinium ester, a pyridopyridazine, a dioxerane, a bioluminescent protein and a luciferase.

23. (Previously presented) The method of claim 3 wherein the second antibody is labelled with a label selected from the group consisting of a metal complex, a stable free radical, a vesicle, a liposome, a colloidal particle, a latex particle, a spin label, biotin and avidin.

24. (Previously presented) The method of claim 6, wherein the solid support is selected from the group consisting of an ELISA plate, a polyacrylamide matrix, a polystyrene tube, polystyrene beads, latex particles, paramagnetic particles, acrylic particles and gelatin particles.

25-33. (Canceled).